

IN THE CLAIMS:

1. (currently amended) A method for the determination of lipid individual molecular species composition of matter ~~and amount by algorithm analysis~~ in a biological sample, said method comprising:

subjecting the biological sample to lipid extraction to obtain a lipid ~~extract and~~ extract;

subjecting the lipid extract to two dimensional ~~(or multidimensional)~~ ~~(or multi-dimensional)~~ electrospray ionization tandem mass spectrometry (ESI/MS/MS) ~~by iterative processing producing the determination of structure and amount~~ to generate a two dimensional plot representing molecular ions of the lipid extract on a first axis and at least one of neutral loss scans of fatty acids of the lipid extract and precursor ion scans on a second axis; and

comparing peak heights for the molecular ions with that for an internal standard to at least one of identify and quantify the lipid molecular species.

2. (currently amended) A method in accordance with Claim 1 wherein the lipid ~~extraction~~ extract is obtained via at least one of a chloroform lipid extraction, a chloroform/methanol extraction, and a butanol extraction.

3. (currently amended) A method in accordance with ~~Claim 2~~ Claim 1 wherein said extraction is of at least one of a blood, serum, tissue, tissue biopsy, feces and urine sample.

4. (currently amended) A method in accordance with ~~Claim 3~~ Claim 1 wherein said ~~[[TG]]~~ biological sample is at least one of a mammalian tissue and a tissue, a plant tissue, a microbiological sample, and a fungal sample.

5. (currently amended) A method in accordance with Claim 4 wherein the mammalian tissue is human tissue and the lipid is at least one of a triacylglyceride, a phospholipid, and any other lipid species contained within biologic membranes.

6. (currently amended) A method in accordance with ~~Claim 5~~ wherein the ~~determination comprises a finger print~~ Claim 1 further comprising determining a fingerprint profile of a ~~patient's triglyceride molecular species and provides a quantitative analysis of individual species~~ lipid individual molecular species.

7. (currently amended) A method in accordance with Claim 6 wherein said ~~finger fingerprint~~ profile ~~comprises~~ represents the individual molecular species of a triglyceride lipid composition of matter.

8. (currently amended) A method for the determination of lipid individual molecular species composition of matter directly from a lipid extract of a biological sample, said method comprising:

subjecting said lipid extract to electrospray ionization tandem mass spectrometry to generate a two dimensional plot of molecular ions of the lipid extract versus at least one of neutral loss scans and precursor ion scans of lipid classes of the lipid extract; and

comparing peak heights for the molecular ions with that for an internal standard to identify and/or quantify the lipid molecular species.

9. (currently amended) A method in accordance with ~~Claim 1~~ Claim 8 wherein said lipid ~~extraction~~ extract is obtained via at least one of chloroform extraction, a chloroform/ methanol extraction, and a butanol extraction.

10. (currently amended) A method in accordance with ~~Claim 9~~ Claim 8 wherein said ~~[[TG]]~~ biological sample is at least one of a mammalian ~~[[or]]~~ and a plant tissue.

11. (original) A method in accordance with Claim 10 wherein said mammalian tissue is human tissue.

12. (currently amended) A method in accordance with ~~Claim 6~~ Claim 8 wherein the biological sample is an aqueous human fluid sample subjected to at least one of centrifugation and/or and conventional column chromatography suitable for separation of lipoproteins to resolve lipids into different lipoprotein fractions.

13. (currently amended) A method in accordance with ~~Claim 6~~ Claim 12 wherein the aqueous human fluid sample is ~~selected from the group consisting of~~ at least one of whole blood, blood serum, blood plasma, liver and urine.

14. (currently amended) A method in accordance with Claim 13 wherein the lipid extract is obtained by extraction of said biological sample with at least one of chloroform and any other solvent.

15. (currently amended) A method in accordance with ~~Claim 14~~ wherein the triglyceride molecular species of the biological sample are determined by comparison with the triglyceride molecular species of a standard Claim 8 wherein said internal standard includes a control sample of lipid molecular species.

16. (currently amended) A method in accordance with ~~Claim 15~~ wherein the triacylglyceride molecular species of the biological sample are determined by comparisons of their ion peak intensities with the ion peak intensities of a standard control sample and iteratively deconvoluted and optionally normalized to yield quantitative information on mass of molecular species. Claim 8 further comprising at least one of iteratively deconvoluting and normalizing the peak heights for the molecular ions.

17. (currently amended) A method in accordance with ~~Claim 16~~ wherein said determination includes deconvolution of Claim 8 further comprising deconvoluting the intensity of two dimensional (or multidimensional) or multidimensional intercept contours of the triglycerides at their neutral loss products at least one of the neutral loss scans and the precursor ion scans for multidimensional mass spectrometry.

18. (original) A diagnostic kit for the determination of lipid molecular species in a biological sample comprising components suitable for carrying out any of the method of Claim 1 and quantitative determinations described therein.

19. (currently amended) A method for assessing a risk to an individual based on [[TG]] molecular species as an independent factor in the development of at least one condition in that individual for a medical condition selected from coronary artery disease,

diabetes, stroke, atherosclerosis and obesity which comprises obesity, wherein the method comprises:

analyzing a biological sample taken of an individual for [[TG]] molecular species determination[[,]] by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of at least one of fatty acids and lipid classes that include constituents present in the lipid extract;

administering a therapeutic amount of a drug to the ~~individual (treated)~~, individual;

analyzing a corresponding biological sample of the ~~treated individual~~, individual;

comparing the [[TG]]molecular species determination after drug administration with the [[TG]]molecular species determination prior to the drug administration; and

determining the benefit of decreased risk due to the drug now afforded to that individual.

20. (currently amended) A method in accordance with Claim 19 wherein the comparison of the [[TG]] molecular species determination of the biological samples is indicative of development of the condition for that individual.

21. (currently amended) A method for identifying an agent which selectively targets specific to at least one of a lipid [[or]] and triacylglyceride molecular species (e.g., saturated triacylglycerides) ~~which comprises species, wherein the method comprises:~~

analyzing a biological sample of at least one treated individual for [[TG]] molecular species or lipid molecular species determination[[,]] by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of the lipid extract;

administering a drug to the ~~individual~~, individual;

analyzing a biological sample from ~~said treated to individual~~, the individual;

comparing the [[TG]] molecular species determination after said administration with the [[TG]] molecular species determination prior to drug administration; and

determining an effect on the ~~treated~~ individual of the drug administration.

22. (currently amended) A method in accordance with Claim 21 wherein said comparison of the [[TG]] molecular species determination of the biological samples is indicative of development of the condition for that individual.

23. (currently amended) A method of identifying a candidate lipid modulating drug having lipid modulating drug ~~efficacy which comprises~~ efficacy, wherein the method comprises:

analyzing a biological sample of at least one individual subject for [[TG]] molecular species determination[, ] by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of the lipid extract;

administering a therapeutic amount of a candidate lipid modulating drug to the individual ~~subject,~~ subject;

analyzing a biological sample of said ~~administered individual,~~ the individual;

comparing the [[TG]] molecular species determination after said administration with the [[TG]] molecular species determination prior to the drug administration; and

determining an effect if any on the individual of the drug administration.

24. (currently amended) A method in accordance with Claim 23 wherein said comparison of [[TG]] analysis is indicative of a lipid modulating capacity of an administered drug.

25. (currently amended) A method in accordance with Claim 24 wherein said modulating comprises lowering.

26. (currently amended) A method for diagnosing and determining the response of a patient to tailored drug ~~therapy which comprises~~ therapy, wherein the method comprises:

analyzing a biological sample of a patient to be treated or ~~[[TG]]~~ or other lipid molecular species determination~~[[,]]~~ by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of the lipid extract;

administering an amount of a drug to the ~~patient;~~ patient;

analyzing a biological sample taken from the treated ~~patient;~~ patient;

comparing the ~~[[TG]]~~ molecular species determination after the administration with the ~~[[TG]]~~ molecular species determination prior to the drug administration; and

determining an effect on the ~~treated~~ patient of the drug administration.

27. (currently amended) A method in accordance with Claim 26 wherein said comparison of ~~[[TG]]~~ analysis is indicative of a successful tailored drug therapy.

28. (currently amended) A method of screening candidate chemicals for lipid modulating efficacy in a ~~subject which comprises~~ subject, wherein the method comprises:

analyzing a biological sample of a subject for ~~[[TG]]~~ molecular species determination~~[[,]]~~ by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of the lipid extract;

administering a therapeutic amount of a drug to that biological ~~subject;~~ subject;

analyzing a biological sample of said ~~subject;~~ subject;

comparing the ~~[[TG]]~~ molecular species determination after said administration with the ~~[[TG]]~~ molecular species determination prior to the drug administration; and

determining an effect if any on the subject of the drug administration.

29. (currently amended) A method of screening in accordance with Claim 28 wherein said comparison of [[TG]] analysis is indicative of a candidate chemical having a lipid lowering potential on a human subject.

30. (currently amended) A method of treating a subject comprising analyzing a biological sample taken of that subject for lipid ~~i.e. TG~~ molecular species determination by the method by multidimensional ESI/MS and quantitative changes, wherein the biological sample is analyzed by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of the lipid extract.

31. (original) A medical treatment in accordance with Claim 30 wherein the subject is a living human.

32. (original) A medical treatment in accordance with Claim 31 wherein said treatment is medicinal and therapeutic.

33. (currently amended) A medical treatment comprising analyzing a biological sample taken of a subject for [[TG]] molecular analysis determination by multidimensional ESIMS and prescribing a therapy based on the determination, wherein the biological sample is analyzed by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of the lipid extract.

34. (original) A medical treatment in accordance with Claim 33 wherein the subject is a human.

35. (original) A method in accordance with Claim 34 wherein said medical treatment is therapeutic.

36. (currently amended) A method of customizing a drug therapy lipid ~~i.e. for~~ a ~~subject which comprises~~ subject, wherein the method comprises:

analyzing a biological sample taken of the subject for [[TG]] molecular species determination by multidimensional ESI/MS by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of at least one of fatty acids and lipid classes that include constituents present in the lipid extract; and

customizing the subject's drug therapy based on the results of the [[TG]] molecular species determination and quantitative changes.

37. (currently amended) A method of customizing drug therapy in accordance with Claim [[34]] 36 wherein the subject is human.

38. (currently amended) A method of retarding, preventing, ameliorating or diagnosing disease in a subject based on lipid ~~i.e. TG~~ molecular species determination of a biologic sample of the subject, ~~which comprises~~ wherein the method comprises:

analyzing a biological sample taken of a subject for [[TG]] molecular species analysis by multidimensional ESIMS determination associated with the disease by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of at least one of fatty acids and lipid classes that include constituents present in the lipid extract; and

prescribing a therapy for the subject based on the alterations in the molecular species profiles of [[TG]] or other lipid molecular species after determination by multidimensional mass spectrometry.

39. (original) A method in accordance with Claim 38 wherein the subject is human.

40. (currently amended) A method of managing a library of ~~chemicals which comprises~~ chemicals, wherein the method comprises:

administering a chemical selected from the library to a ~~subject and~~ subject;



analyzing a biological sample taken of that subject for lipid ~~i.e.~~ TG molecular species determination by multidimensional ESI/MS[.,.] by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of at least one of fatty acids and lipid classes that include constituents present in the lipid extract;

quantitating the mass of individual entities; and

assigning a priority to said chemical for further development based on that determination.

41. (original) A method in accordance with Claim 40 wherein the subject is human.

42. (currently amended) A method of determining a subject's response to administration of a ~~drug which comprises~~ drug, wherein the method comprises:

administering a drug to the ~~subject,~~ subject; and

analyzing a biological sample taken of a subject for lipid ~~i.e.~~ TG molecular analysis by multidimensional ESI/MS following said administration, molecular species and quantitation, wherein the biological sample is analyzed by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of at least one of fatty acids and lipid classes that include constituents present in the lipid extract.

43. (original) A method in accordance with Claim 42 wherein the subject is human.

44. (currently amended) A method of providing a medical assessment to a ~~subject which comprises~~ subject, wherein the method comprises:

analyzing a biological sample taken of a subject for lipid ~~i.e.~~ TG molecular analysis by multidimensional ESI/MS by comparing peaks heights of a plot of molecular ions of a lipid extract of the molccular species versus at least one of neutral loss scans and precursor

ion scans of at least one of fatty acids and lipid classes that include constituents present in the lipid extract; and

providing an assessment to the subject based on that determination.

45. (original) A method of providing a medical assessment in accordance with Claim 44 wherein the subject is human.

46. (currently amended) A method of enhancing medical care provided to a ~~subject~~ which comprises subject, wherein the method comprises:

analyzing a biological sample taken of a subject for [[TG]] or other lipid molecular analysis by multidimensional ESI/MS by comparing peaks heights of a plot of molecular ions of a lipid extract of the molecular species versus at least one of neutral loss scans and precursor ion scans of at least one of fatty acids and lipid classes that include constituents present in the lipid extract; and

providing a modulated therapy to the subject.

47. (original) A method of enhancing subject care in accordance with Claim 46 wherein the subject is human.

48. (currently amended) A method in accordance with Claim 47 wherein the [[TG]] molecular analysis is a [[TG]] molecular analysis of species and simultaneous quantitation.

49-54. (canceled)

55. (original) A method in accordance with Claim 1 wherein said lipid comprises at least one of phospholipids (e.g., choline glycerophospholipides (e.g., plasmenycholine, phosphatidylcholine, plasmanylcholine), sphingomyelin, ethanolamine glycerophospholipids, mono and dimethyl ethanolamine, glycerophospholipids, serine glycerophospholipids, inositol glycerophospholipids, cardiolipin, phosphatidic acid, phosphatidylglycerol, phosphatidylethanol and oxidized derivatives thereof), fatty acids, fatty amides, eicosanoids, sphingolipids, glycolipids, steroids, ceramides, acylCoA, acylcarnitine,

acylproteins, acylpeptides, diglycerides, monoglycerides, anadamide and 2-arachidonyl glycerol or oxidized nitrated or sulfated species therefrom or other derivatives known to those in the field.

56. (original) A method in accordance with ~~Claims 19, 20 and 23~~ Claim 19 wherein said lipid comprises at least one of phospholipids (e.g., choline glycerophospholipids (e.g., plasmalogen, phosphatidylcholine, plasmalogen), sphingomyelin, ethanolamine glycerophospholipids, mono and dimethyl ethanolamine, glycerophospholipids, serine glycerophospholipids, inositol glycerophospholipids, cardiolipin, phosphatidic acid, phosphatidylglycerol, phosphatidylethanol and oxidized derivatives thereof), fatty acids, fatty amides, eicosanoids, sphingolipids, glycolipids, steroids, ceramides, acylCoA, acylcarnitine, acylproteins, acylpeptides, diglycerides, monoglycerides, anadamide and 2-arachidonyl glycerol.